Phase II Study of Perioperative Chemotherapy with Cisplatin and Pemetrexed in Non–Small-Cell Lung Cancer

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Introduction: Pathologic complete response (pCR) with neoadjuvant chemotherapy is associated with improved survival in many solid tumors. We evaluated pCR rate of cisplatin with pemetrexed in non–small-cell lung cancer.

Methods: Patients with stages IB to IIIA non–small-cell lung cancer, Eastern Cooperative Oncology Group performance status 0 to 1 were enrolled in this single-arm phase II trial using two-stage design with 90% power to detect pCR rate of more than or equal to 10%. Pretreatment mediastinal lymph node biopsy was required. Patients received three cycles of cisplatin 75 mg/m^2 with pemetrexed 500 mg/m^2 (day 1 every 21 days) preoperatively and additional two cycles within 60 to 80 days after surgery. The primary end point was pCR. Polymorphisms in *FPGS, GGH*, *SLC19A1*, and *TYMS* genes were correlated with treatment outcomes.

Results: Thirty-eight patients were enrolled, with median age of 62.5 years. Preoperatively, 26% had squamous histology, and 34% had biopsy-proven N2 involvement. R0 resection was achieved in 94% of the 34 patients who underwent surgery, and 54% had documented N2 clearance. There was no pCR seen. Median disease-free survival (DFS) and overall survival of these patients have not yet been reached in contrast to median of 13.8 and 24.2 months, respectively, in patients with persistent N2 disease ($p = 0.3241$ and $p = 0.1022$, respectively). There was a statistically significant association between DFS and postoperative tumor, node, metastasis stage ($p = 0.0429$), *SLC19A1* rs3788189 TT genotype ($p = 0.0821$), and viable tumor defined as less than or equal to 10% of resected specimen ($p = 0.026$).

Conclusion: The primary end point was not met. Patients with N2 clearance, less than or equal to 10% viable tumor in the resected specimen, and *SLC19A1* rs3788189 TT genotype have favorable DFS outcomes.

Key Words: Neoadjuvant chemotherapy, Non–small-cell lung cancer, Pemetrexed, Cisplatin, Pharmacogenetics.

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Lung cancer is the leading cause of cancer deaths in the United States, with estimated 228,080 new cases and 159,480 deaths in 2013.[1] Although surgery is curative for resectable stages of non–small-cell lung cancer (NSCLC), 5-year survival estimates are inferior to breast, colon, and prostate cancer. As distant relapse is the predominant reason for treatment failure upon long-term follow-up, this has led to the investigation of the role of systemic therapy in the perioperative setting. Delivering chemotherapy preoperatively offers the potential for control of micrometastatic disease and tumor debulking to reduce the extent of planned surgery. Moreover, a few randomized studies in the 1990s demonstrated survival improvements with preoperative chemotherapy compared with surgery alone.[2–4] In contrast to the lack of survival benefit for patients with pathologic complete response (pCR) after receiving preoperative radiation in NSCLC,[5,6] pCR seen after preoperative chemotherapy is associated with improved survival outcomes and is a major end point in the evaluation of preoperative chemotherapy trials.[7–9]

In 2003–2004, several large phase III trials confirmed a significant benefit from adjuvant chemotherapy in the treatment of early-stage NSCLC.[10–13] In addition, the meta-analysis "Lung Adjuvant Cisplatin Evaluation" confirmed a survival benefit from cisplatin-based adjuvant chemotherapy in lung cancer.[14] It is important to note, however, that sustained survival benefit is not all encompassing and dependent on the chemotherapy regimen utilized.[15]

Based on the high systemic failure rates even in completely resected NSCLC, we designed a phase II trial evaluating cisplatin in combination with pemetrexed administered neoadjuvantly followed by adjuvant treatment for surgical candidates with stage IB to IIIA NSCLC. Pemetrexed is metabolized by the folate pathway enzymes, including folylpolyglutamate synthetase (FPGS), γ-glutamyl hydrolase, solute carrier 19A1 (SLC19A1), and thymidylate synthase (TYMS). Polymorphisms in the genes encoding these proteins have been associated with variable treatment outcomes[16–21]; therefore, we investigated the correlation between single-nucleotide polymorphisms (SNPs) in these four genes and the clinical outcomes for this study.

PATIENTS AND METHODS

Eligibility

Eligible patients must have had newly diagnosed NSCLC, disease stage T2N0M0, T1-3N1M0, or T1-3N2M0

(as defined by 6th edition of TNM classification) and deemed to be potential surgical candidates by a thoracic surgeon. Patients were required to have adequate bone marrow, hepatic, and renal function (creatinine clearance ≥45 ml/min based on the Cockcroft and Gault formula) and Eastern Cooperative Oncology Group performance status 0 to 1. Patients with distant metastasis, biopsy-proven N3 nodal involvement, malignant pleural effusion, or a history of other malignancy within the preceding 2 years (except nonmelanoma skin cancer and cervical carcinoma in situ) were excluded from this trial. Subsequent to the findings released in 2008–2009 regarding inferior outcomes with pemetrexed treatment among patients with squamous cell NSCLC,[22,23] patients with predominant squamous cell carcinoma histology were excluded starting January 2010. The trial was approved by the Roswell Park Cancer Institute Institutional Review Board, and each patient provided written informed consent before starting protocol-related activities.

Study Treatment and Procedures

All patients underwent chest and abdomen computed tomography (CT) scan for tumor measurement, positron emission tomography (PET) scan, electrocardiography, and pulmonary function tests for screening at baseline. Mediastinal lymph node evaluation was performed with either mediastinoscopy or endobronchial ultrasound-guided biopsy of mediastinal lymph nodes before initiating protocol treatment. Patients were given vitamin B12 (repeated once every 9 wk) and folic acid supplementation at least 1 week before cycle 1 day 1 of chemotherapy. Cisplatin 75 mg/m^2 and pemetrexed 500 mg/m^2 were administered on day 1 of each 21-day cycle along with standard hydration and antiemetic regimens. They were evaluated weekly using the National Cancer Institute Common Toxicity Criteria (CTC) version 3.0 to assess severity of adverse effects. After three cycles of chemotherapy, CT chest and abdomen and PET scans were repeated. Response Evaluation Criteria in Solid Tumors (RECIST) criteria version 1.0 was used for evaluation of objective response rate (ORR).

Patients with complete response, partial response, or stable disease by CT and PET scans underwent surgery, no earlier than 28 days after the last dose of chemotherapy but between days 70 to 90 from the start of treatment. Patients underwent either standard thoracotomy or video-assisted thoracoscopic surgery for a lobectomy, bilobectomy, or pneumonectomy with mediastinal lymph node dissection as described by Martini.[24]

Patients deemed to have unresectable disease or disease progression were subsequently removed from study treatment and followed every 6 months until the time of death. Patients who had R0 resection, with an Eastern Cooperative Oncology Group performance status of 0 to 1, received two additional cycles of cisplatin with pemetrexed within 60 days after surgery provided there were no uncontrolled intercurrent illnesses. Patients with incomplete resections, positive margins or those found to have multistation N2 disease or extracapsular lymph nodal involvement could receive adjuvant radiation following adjuvant chemotherapy according to standard of care. Follow-up visits with surveillance imaging studies were performed every 3 months for 2 years, then every 6 months for 3 years, and annually thereafter.

Study Design

A two-stage phase II study based on Simon design was conducted to assess the efficacy of this regimen. The primary end point was pCR, which was defined as surgical pathology specimen free of all gross and microscopic evidence of viable tumor. Patients who consented to the study but did not receive any chemotherapy were excluded from the analysis. Patients were considered evaluable for determination of primary end point if they received at least one cycle of preoperative chemotherapy. For the purpose of the decision rule, in addition to the pCR definition, the following were considered equivalent to not having a pCR: interval development of radiologic findings that contraindicate surgery, surgery being aborted according to the best judgment of the surgeon, R1 or R2 resection. For descriptive purposes only, overall and disease-free survival (DFS) estimates were computed using the Kaplan–Meier method. Survival outcomes were calculated from the date of administration of first chemotherapy cycle.

The trial was designed to test the null hypothesis that the probability of pCR was less than 2% against the alternative more than 10%, using a sample size of 52 with a significance level of 0.08 and power of 90%. To test this hypothesis, 38 patients were to be accrued during the first stage. Accrual was completed at this stage if either zero or at least three pCRs were documented and the regimen considered inactive or active, respectively. If at least two pCRs were observed, study would accrue an additional 14 patients in the second stage. If no additional pCRs were noted, regimen would be considered inactive. If there were at least three documented pCRs among the 58 patients accrued, the regimen would be considered active.

Correlative Pharmacogenetic Studies

Ten milliliters of blood was drawn into ethylenediaminetetraacetic acid tubes at baseline and DNA extracted and preserved until the end of the study for genotyping. SNP information for *FPGS*, *γ-glutamyl hydrolase*, *SLC19A1*, and *TYMS* were acquired as previously described and had also been identified in preliminary studies as potentially prognostic or predictive for treatment outcomes with pemetrexed.[19–21] Genotyping of the SNPs has been described elsewhere.[20]

Statistical Analysis

As the 7th edition of TNM staging was proposed and published in 2007, the data obtained initially using the 6th edition were reclassified as appropriate. Descriptive statistics such as frequencies and relative frequencies were computed for categorical variables. Numeric variables were summarized using simple descriptive statistics, such as the mean, SD, median, and range. Fisher's exact test was used to study the association between categorical variables. Adverse events were tabulated and reported using the CTC version 3.0. The Wilcoxon ranked sum test was used to compare the groups with regard to numeric variables. The estimated overall survival (OS) and DFS distributions were obtained

using the Kaplan–Meier method. Using this distributional estimate, summary descriptive statistics such as the median survival and a 95% confidence interval of the median survival were obtained. Statistical assessment of observed differences in the survival distributions of different groups of interest was done using the log-rank test. The Cox proportional hazards model was used to assess the association between numeric variables and the survival. Exact 95% confidence interval using the Clopper–Pearson method was obtained for the ORR. SNP analysis was performed using dominant model, that is, the homozygous alleles with lower prevalence were combined with the heterozygous alleles versus the homozygous alleles with higher prevalence. Adjusted *p* values were also provided using the linear step-up method of Benjamini and Hochberg. A 0.05 nominal significance level was used in all testing. All statistical analyses were done using SAS (version 9.3; SAS Institute, Cary, NC).

RESULTS

Patients

From September 2005 to January 2011, 38 patients were accrued. Baseline patient characteristics are summarized in Table 1. Male to female ratio was 1:1. Adenocarcinoma was the most frequent histology (55%). Thirty-two patients had already been accrued before the protocol amendment excluding squamous cell histology by January 2010. Nineteen patients (50%) had clinical stage IIIA disease. Thirteen patients had biopsy-proven N2 nodal involvement before treatment. Due to logistical challenges (such as PET scan performed at another institution), baseline and postchemotherapy PET scan maximal standardized uptake values (SUV) were available for analysis in 16 patients only.

TABLE 1. Baseline Characteristics of 38 Patients Enrolled

Variable	Results (%)
Age	
Median	62.5
Range	39–75
Sex	
Male	19 (50%)
Performance status	
0	28 (74%)
1	10 (26%)
Histology	
Squamous cell	10 (26%)
Adenocarcinoma	21 (55%)
Large-cell neuroendocrine	3 (8%)
Poorly differentiated, not otherwise specified	4 (11%)
Clinical stage (preoperative)	
IB	5 (13%)
IIA	7 (18.5%)
IIB	7 (18.5%)
IIIA	19 (50%)
Biopsy-proven N2 nodal involvement before initiation of chemotherapy	13 (34%)

Systemic Treatment and Toxicities

All patients received at least one cycle of chemotherapy. Thirty-five patients (92%) received all three induction treatment cycles. A median of four cycles of chemotherapy (including adjuvant) was administered. Eighteen patients (47%) received five cycles without dose reductions or dose delays. One patient had both dose reduction and dose delay (3%). Among three patients who received less than three cycles of neoadjuvant chemotherapy, two patients discontinued therapy due to toxicity (one for CTC grade 3 drug fever and the other for CTC grade 3 sensory neuropathy) and the third patient was taken off protocol due to interval disease progression. CTC more than or equal to grade 3 nonhematologic adverse events (<10%) included arterial thrombosis, bronchopleural fistula, empyema, hyperglycemia, fatigue, and tinnitus. There were no CTC grade 5 events attributable to study treatment. Table 2 lists frequency of relevant toxicities encountered in this study.

Treatment Response according to Imaging Modality

The ORR for all 38 patients was 29% (11 partial responses). Disease progression was noted in 11% ($n = 4$). Stable disease was noted in 60% ($n = 23$). There was no radiologic complete response noted. There was no association between ORR with histologic type, baseline, or postchemotherapy PET scan SUV value or absolute change of PET scan SUV value before and after chemotherapy. Percent change of maximal SUV between baseline and postchemotherapy PET scan correlated with ORR ($p = 0.0259$), with median percent SUV reduction among responders of 68.5% (range, 57.1%–78.7% reduction) compared with median of 29.5% reduction among nonresponders (ranging between increase in percent SUV of 51.5% to reduction of 92.4%). Nevertheless, as described previously, complete data were only available in 16 of 38 patients.

Surgical Treatment, Morbidity, and Mortality

Of 38 patients, 34 patients (89%) underwent thoracotomy. Four patients did not undergo surgery due to the following reasons: Three patients had disease progression by RECIST criteria which precluded surgery. One patient who had stable disease radiographically after induction chemotherapy had hypoxia due to difficult intubation process and surgery was aborted.

Among the 34 patients who underwent were able to undergo surgery, 32 patients (94%) had R0 resection. Median number of resected lymph nodes (including the ones sampled through mediastinoscopy before starting neoadjuvant chemotherapy) was 14.5 (range, 5–42). Complete resection was achieved in 23 patients (67.6%) by thoracoscopic approach. Nine patients (26.5%) underwent conversion to standard thoracotomy after initial video assisted thoracoscopic surgery (VATS) approach, whereas two patients underwent standard thoracotomy upfront (5.9%). Six patients (17.6%) underwent pneumonectomy, four of which were completed thoracoscopically. All remaining resections were lobectomies with exceptions for the two R2 resection cases as described below.

TABLE 2. Adverse Events Encountered per Patient Attributed to Chemotherapy

	All CTC Grades		CTC Grade 3		CTC Grade 4	
Adverse Event	***n***	**%**	***n***	**%**	***n***	**%**
Fatigue	22	57.9	3	7.9	0	0
Nausea	22	57.9	0	0	0	0
Anemia	19	50	1	2.6	0	0
Diarrhea	12	31.6	0	0	0	0
Tinnitus	10	26.3	1	2.6	0	0
Neutropenia	10	26.3	2	5.3	0	0
Vomiting	7	18.4	0	0	0	0
Sensory neuropathy	6	15.8	1	2.6	0	0
Elevated transaminase	5	13.2	0	0	0	0
Thrombocytopenia	5	13.2	0	0	0	0
Hyperglycemia	4	10.5	2	5.3	0	0
Constipation	3	7.9	0	0	0	0
Arterial thromboembolism	1	2.6	1	2.6	0	0
Empyema	1	2.6	1	2.6	0	0
Bronchopleural fistula	1	2.6	1	2.6	0	0
Febrile neutropenia	0	0	0	0	0	0

CTC, Common Toxicity Criteria.

One of the two patients classified to have R2 resection had disease progression by RECIST due to interval development of separate ipsilateral left upper lung nodules despite mild reduction in size of the primary left lower lobe hilar mass. This patient underwent planned VATS wedge excision of one of the new nodules which was confirmed intraoperatively to be malignant (T4 using the current 7th edition of TNM staging). Further surgery was not pursued. This patient was classified as having R2 resection and received a different chemotherapy regimen subsequently. The second patient with R2 resection had stable disease by RECIST on the post-chemotherapy imaging studies but underwent VATS exploratory thoracoscopy only as there was a pleural nodule found intraoperatively separate from the tumor mass. Frozen sections of this nodule and another N2 node documented presence of malignant cells.

Perioperative mortality rate in this small cohort was 0%. Four patients required reoperation or bronchoscopic evaluation within 30 days for the following reasons: One patient had persistent air leak requiring suture repair of bronchopleural fistula and mechanical pleurodesis 8 days after initial VATS left upper lobectomy. Another patient required emergent thoracoscopy for evacuation of hematoma 3 days after surgery. A third patient underwent decortication and drainage of empyema 19 days after the preceding thoracotomy. The last patient had mucus plugging causing lobar collapse for which the patient underwent bronchoscopic evaluation and suctioning. A total of four patients required rehospitalization within 30 days for the following reasons: postoperative ileus 2 days after initial hospital discharge (postoperative day 4), bilateral bronchopneumonia 2 days after initial hospital discharge (postoperative day 4), symptomatic worsening dyspnea 10 days after hospital discharge (postoperative day 12) requiring inpatient pulmonary toilet, and one patient with empyema who was readmitted to the hospital (postoperative day 6) 1 day after initial hospital discharge who then underwent decortication as described above.

Pathologic Assessment

Pathologic response of the lung cancer to induction chemotherapy was evaluated on standard hematoxylin–eosin histopathology on the 33 patients who underwent at least a wedge resection of a lung tumor specimen. Proportion of viable tumor, necrosis, and fibrosis/inflammation were each determined in increments of 5% for each specimen (composite total of 100%). There was no pCR noted during the first stage and thus accrual to the second stage was not pursued. Percent viable tumor ranged from 5% to 100%, with median of 70% viable residual tumor. Eight patients, of whom five had partial response (PR) by RECIST criteria, had less than or equal to 10% viable malignant cells in the resected specimen (four patients with 5% and four patients with 10% viable tumor remaining). Percentage necrosis ranged from 0% to 30%, with a median of 10% necrosis (0% in 11 patients, 5% in two patients, 10% in six patients, 15% in one patient, 20% in nine patients, and 30% in four patients). Percentage of fibrosis/inflammation ranged from 0% to 95%, with a median of 20% fibrosis/inflammation (<50% in 21 patients and ≥50% in 12 patients).

Seventeen patients had suspected N2 nodal involvement based on CT and/or PET criteria before initiation of chemotherapy. Thirteen patients had biopsy-proven N2 nodal involvement before chemotherapy. Eleven of these 13 patients underwent thoracotomy, and N2 nodes were positive in four patients and negative in seven patients. One patient who had pretreatment PET-positive paratracheal adenopathy that was negative by mediastinoscopy performed before starting chemotherapy and clinically negative N2 involvement by CT/PET criteria after three cycles of chemotherapy before

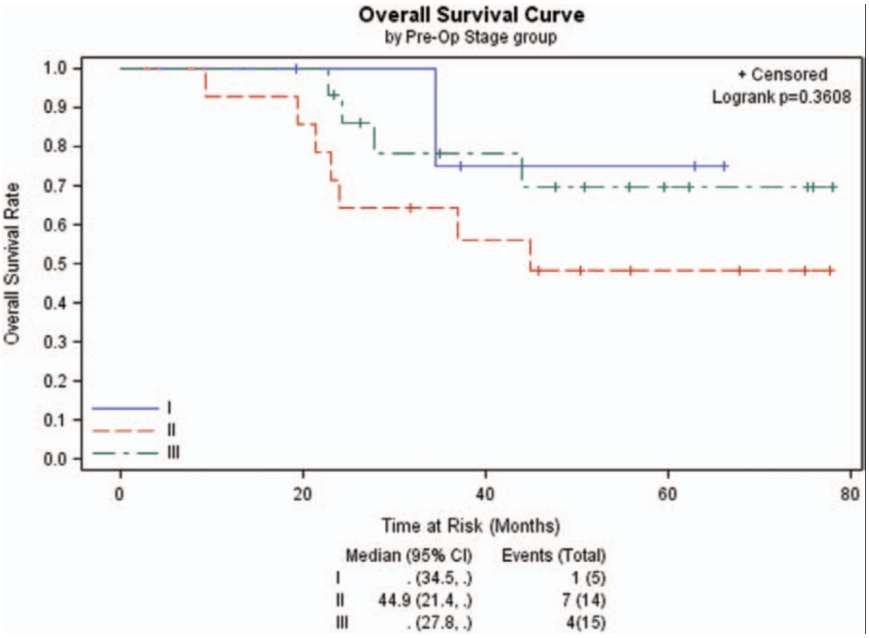

FIGURE 1. Overall survival Kaplan–Meier curve according to preoperative stage (n = 34). CI, confidence interval.

surgery was found to have microscopic involvement of three resected subcarinal lymph nodes in the permanent hematoxylin–eosin sections after lobectomy. Thus, a total of five patients had N2 lymph node involvement documented in their postchemotherapy surgical specimen. Two of these five patients received postoperative radiation following additional chemotherapy within 4 months after surgery.

Survival

As of November 30, 2012, median follow-up was 37.1 months (range, 1.5–78 mo). Disease progression had occurred in 20 patients (52.6%). Documented relapses were distant except in two patients: one patient had biopsy-proven mediastinal lymph node involvement as the solitary site of relapse 14 months after initial surgery. He received concurrent chemoradiation but subsequently died 28 months thereafter from cardiac complications. The second occurred in the patient who underwent definitive chemoradiation after surgery was aborted due to hypoxia during intubation as described above. Surveillance imaging showed locoregional disease progression 16 months after completing chemoradiotherapy. He opted for best supportive care only with no further imaging studies and died 15 months later. Figure 1 shows the OS curve of clinical tumor, node, metastasis grouping preoperatively in the 34 patients who underwent surgery. Figures 2 and 3 show the Kaplan–Meier DFS and OS curves according to postoperative

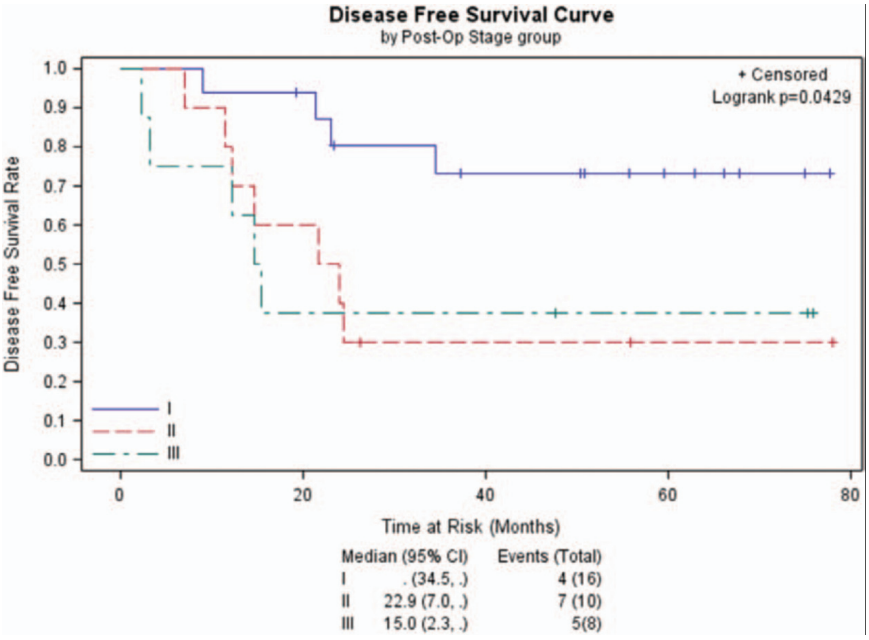

FIGURE 2. Disease-free survival Kaplan–Meier curve according to postoperative stage (n = 34). CI, confidence interval.

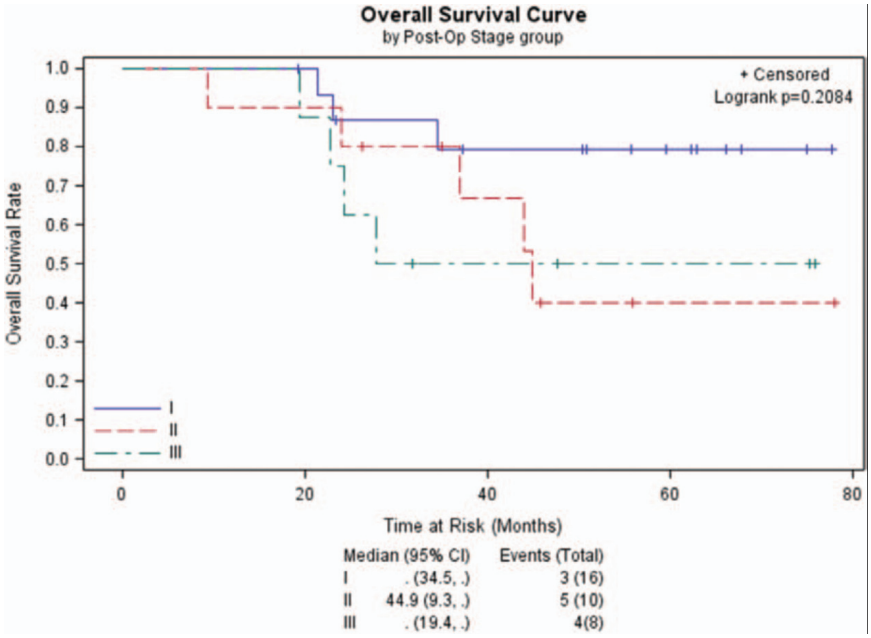

FIGURE 3. Overall survival Kaplan–Meier curves according to postoperative stage (n = 34). CI, confidence interval.

pathologic stage. For the entire cohort, median DFS was 24.5 months (95% CI, 15.4, not reached), with 2-year and 5-year DFS rate of 54.8% (95% CI, 37.7%, 69%) and 46% (95% CI, 29.4%, 61.1%), respectively. Median OS for the entire cohort, as shown in Figure 4, has not been reached (95% CI, 35.9, not reached), with 2-year and 5-year OS rate of 81% (95% CI, 64.2%, 90.5%) and 55.6% (95% CI, 37%, 70.6%), respectively.

Variables Associated with Survival Outcomes

Among patients who underwent surgery, postoperative stage was the most important variable associated with DFS (p = 0.0429). Nevertheless, this was not significantly associated with OS (p = 0.2084). Figures 2 and 3 show the Kaplan–Meier curves demonstrating DFS and OS according to postoperative stage. Among the patients with N2 nodal clearance, median DFS and OS of these patients have not yet been reached in contrast to 13.8 and 24.2 months, respectively, in patients with persistent N2 disease. The difference is, however, not statistically significant (p = 0.3241 and p = 0.1022, respectively).

Other parameters, such as preoperative stage, objective response, histology, sex, and smoking status, were not associated with either OS or DFS. Although a trend toward improved DFS and OS was observed for those with an objective radiologic response, this difference was not statistically significant

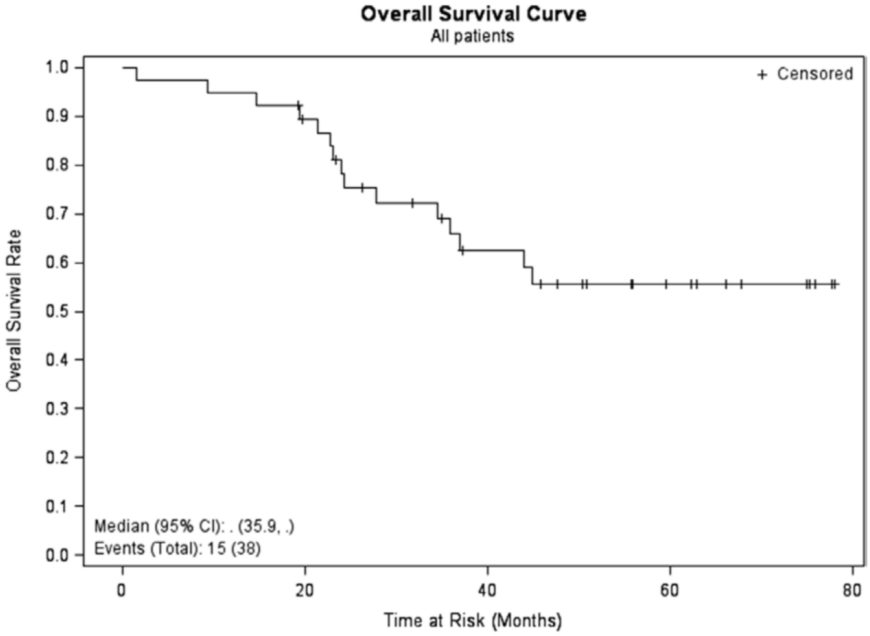

FIGURE 4. Kaplan–Meier curve for overall survival in the entire cohort (n = 38). CI, confidence interval.

(p = 0.3414 and p = 0.4136). Pathologic response as defined by the proportion of viable tumor less than or equal to 10% in the resected specimen was associated with an improved DFS (p = 0.0223) but not OS. When analyzed as continuous variables, percentages of viable tumor or fibrosis/inflammation were associated with DFS (hazard ratio, 1.031; 95% CI, 1.002–1.039; p = 0.026 and hazard ratio, 0.977; 95% CI, 0.956–0.997; p = 0.0282, respectively) but not OS. Percentage of necrosis was associated with neither DFS nor OS. Data for DFS are summarized in Table 3.

Comparing the OS of patients with complete resection (n = 32) with those who had incomplete or no resection (n = 6), we found 2-year and 5-year survival rates of 87.2% (95% CI, 69.4%–95%) and 61.3% (95% CI, 40.7%–76.6%) for the former and 50% (95% CI, 11.1%–80.4%) and 25% (95% CI, 1.2%–64.6%) for the latter patients, respectively. The median OS time for patients with complete resection has not been reached (95% CI, 37% to not reached), and it was 27.7 months (95% CI, 1.5 mo, not reached) for those with incomplete/no resection (p = 0.0136).

Pharmacogenetics

Twenty-one tagSNPs generated from the four genes with minor allele frequency more than 5% were successfully genotyped and were in Hardy–Weinberg equilibrium. Genotypes observed in less than 5% patients were regrouped, and if the regrouped frequency was less than or equal to 10%, the SNP was excluded from the analyses with the clinical outcomes. The SNPs were analyzed for their interaction with ORR, DFS, and OS. Three SNPs correlated with OS and DFS. At the time of analysis, the median survival was significantly decreased for patients with the *FPGS* rs10987742 variant TT genotype (OS: p = 0.0009; DFS: p = 0.0216) and *TYMS* rs2853533 variant CC genotype (OS: p = 0.0005) even though the median OS for *FPGS* rs10987742 CC and *TYMS* rs2853533 GG genotypes has not been reached. *SLC19A1* rs3788189 G>T polymorphism, located in the intronic region of the folate transporter gene, though not statistically significant, was associated with DFS: with the median DFS of 21.6 months for the homozygous GG genotype versus a median DFS that has not yet been reached for the homozygous TT genotype (p = 0.0821). The AA genotype for *SLC19A1* rs2838958 correlated with ORR (trend of association p value = 0.072). Supplementary Tables 1 and 2 (Supplementary Digital Content 1, http://links.lww.com/JTO/A519) list the individual association results for genotypes with ORR, DFS, and OS.

DISCUSSION

Our study is the first phase II study to complete the evaluation of pemetrexed in combination with cisplatin as preoperative chemotherapy followed by adjuvant treatment for stage IB to IIIA NSCLC. Another single-arm phase II study using the same regimen neoadjuvantly for a similar patient population in Spain (NCT002592850) was initiated in December 2005 and projected to enroll 44 patients. This study was, however, terminated in 2008 due to poor accrual and survival outcomes measures were thus not analyzed in the 10 patients who were enrolled. There were five partial responses, one patient with disease progression with a total of seven patients who underwent surgery.[25] A randomized European Organization for Research and Treatment of Cancer (EORTC) phase II study of pemetrexed and cisplatin as either induction or adjuvant chemotherapy in approximately 130 patients with stage IB to II NSCLC was first launched in October 2006 (EORTC-08051,

TABLE 3. Log-Rank Test for Disease-Free Survival in the 34 Patients Who Underwent Surgery

Variable	Level	Total	Number Failed	Number Censored	Median Survival (95% CI)	p
Response	No	23	12	11	24.5 (14.7, NR)	0.3414
	Yes	11	4	7	NR (14.7, NR)	
Histology	Nonsquamous	25	14	11	24.5 (15.4, NR)	0.1159
	Squamous	9	2	7	NR (2.3, NR)	
Preoperative stage	I	5	1	4	NR (34.5, NR)	0.1762
	II	14	9	5	22.4 (7, NR)	
	III	15	6	9	NR (12.2, NR)	
Postoperative stage	I	16	4	12	NR (34.5, NR)	0.0429
	II	10	7	3	22.9 (7, NR)	
	III	8	5	3	15 (2.3, NR)	
Sex	Female	16	8	8	NR (11.5, NR)	0.6719
	Male	18	8	10	NR (15.4, NR)	
Smoking	No	2	1	1	NR (3.2, NR)	0.5177
	Yes	32	15	17	NR (21.4, NR)	
Viable tumor[a]	≤10%	8	1	7	NR (34.5, NR)	0.0223
	>10%	25	15	10	23.1 (14.7, NR)	
Resection	Complete	32	15	17	NR (21.4, NR)	0.6882
	Incomplete	2	1	1	NR (2.3, NR)	

[a]Assessed in 33 of 34 patients who underwent at least a wedge resection of the lung mass. Not included in the analysis was the specimen from one patient who had exploratory thoracoscopy and mediastinal sampling only. Further operation was discontinued when a pleural nodule and a mediastinal lymph node showed presence of malignant cells.

CI, confidence interval; NR, not reached.

NCT00389688). This study also closed in 2008 because of poor recruitment.[26] Although the primary end point was not met, we were able to show that preoperative chemotherapy using this regimen did not result in increased postoperative mortality, similar to what has been reported in larger randomized studies.[27–30] Nevertheless, it is to be emphasized that the standard of care for patients with early-stage NSCLC is surgery upfront followed by adjuvant chemotherapy as indicated.

The pCR rate seen in our study is similar to what has been published for cisplatin-based induction chemotherapy regimens (without the combination of preoperative radiation), ranging typically from 0% to 10%,[27,31] though some studies report pCR rates of 15% to 19%.[32,33] Although this variability is undoubtedly in part due to heterogeneity of NSCLC and type of regimen used, there is lack of uniform or standard definition of pCR across different studies, with some studies indicating more than or equal to 95% necrosis/sclerosis as pCR[33,34] as opposed to the implied definition of complete necrosis or absence of any viable tumor cells. Thus, the presumption that pCR meant a complete absence of cancer cells cannot be taken for granted across studies. In addition, there is a lack of association between histologic type and response rate. Our study did not reveal an inferior outcome among patients with squamous histology. Although this may be related to the small sample size, new findings suggest that gene expression profiling may further refine the ability to identify subset of squamous cell carcinoma who may respond to pemetrexed treatment.[35]

This study is limited by the small number of patients accrued and the lack of a suitable control arm. Although the primary end point was not reached, the results from this study provide further evidence of the strong prognostic factor associated with mediastinal downstaging. A phase II study is currently ongoing, with planned enrollment of 33 patients to evaluate the efficacy of cisplatin and pemetrexed as preoperative chemotherapy for nonsquamous NSCLC patients with N2 nodal involvement (NCT 01165021). Our results support current observations that surgery for selected patients with known N2 disease may result in long-term survival provided there is clear demonstration of pathologic clearance of N2 nodal disease.[33,36] Observations from other retrospective studies suggest that surgery after chemotherapy could still be effective even in patients with persistent N2 disease, provided clinical response was demonstrated and complete resection can be achieved.[31,37,38] Polymorphisms in the folate transporter *SLC19A1* have been shown to be associated with survival outcomes in NSCLC patients treated with pemetrexed-based regimens.[19,20,39] This study revealed that *SLC19A1* rs3788189 polymorphism which was associated with OS in a previous study[19] was also associated with DFS in this study although it did not reach statistical significance. SNPs in *FPGS* and *TYMS* were also found to be associated with inferior OS and/or DFS. Although the functional implication of these polymorphisms is yet to be characterized, in some cases such as the *TYMS* rs2853533 SNP which was associated with inferior OS in this study, it has been linked to 5-FU cytotoxicity.[40]

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REFERENCES

1. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2013. *CA Cancer J Clin* 2012;62:11–30.
2. Depierre A, Milleron B, Moro-Sibilot D, et al; French Thoracic Cooperative Group. Preoperative chemotherapy followed by surgery compared with primary surgery in resectable stage I (except T1N0), II, and IIIa non-small-cell lung cancer. *J Clin Oncol* 2002;20:247–253.
3. Rosell R, Gómez-Codina J, Camps C, et al. Preresectional chemotherapy in stage IIIA non-small-cell lung cancer: a 7-year assessment of a randomized controlled trial. *Lung Cancer* 1999;26:7–14.
4. Roth JA, Fossella F, Komaki R, et al. A randomized trial comparing perioperative chemotherapy and surgery with surgery alone in resectable stage IIIA non-small-cell lung cancer. *J Natl Cancer Inst* 1994;86:673–680.
5. Bromley LL, Szur L. Combined radiotherapy and resection for carcinoma of the bronchus; experiences with 66 patients. *Lancet* 1955;269:937–941.
6. Shields TW, Higgins GA Jr, Lawton R, Heilbrunn A, Keehn RJ. Preoperative x-ray therapy as an adjuvant in the treatment of bronchogenic carcinoma. *J Thorac Cardiovasc Surg* 1970;59:49–61.
7. Elias AD, Skarin AT, Leong T, et al. Neoadjuvant therapy for surgically staged IIIA N2 non-small cell lung cancer (NSCLC). *Lung Cancer* 1997;17:147–161.
8. Martini N, Kris MG, Flehinger BJ, et al. Preoperative chemotherapy for stage IIIa (N2) lung cancer: the Sloan-Kettering experience with 136 patients. *Ann Thorac Surg* 1993;55:1365–1373.
9. Pisters KM, Kris MG, Gralla RJ, Zaman MB, Heelan RT, Martini N. Pathologic complete response in advanced non-small-cell lung cancer following preoperative chemotherapy: implications for the design of future non-small-cell lung cancer combined modality trials. *J Clin Oncol* 1993;11:1757–1762.
10. Strauss GM, Herndon JE 2nd, Maddaus MA, et al. Adjuvant paclitaxel plus carboplatin compared with observation in stage IB non-small-cell lung cancer: CALGB 9633 with the Cancer and Leukemia Group B, Radiation Therapy Oncology Group, and North Central Cancer Treatment Group Study Groups. *J Clin Oncol* 2008;26:5043–5051.
11. Winton T, Livingston R, Johnson D, et al; National Cancer Institute of Canada Clinical Trials Group; National Cancer Institute of the United States Intergroup JBR.10 Trial Investigators. Vinorelbine plus cisplatin vs. observation in resected non-small-cell lung cancer. *N Engl J Med* 2005;352:2589–2597.
12. Arriagada R, Bergman B, Dunant A, Le Chevalier T, Pignon JP, Vansteenkiste J; International Adjuvant Lung Cancer Trial Collaborative Group. Cisplatin-based adjuvant chemotherapy in patients with completely resected non-small-cell lung cancer. *N Engl J Med* 2004;350:351–360.
13. Douillard JY, Rosell R, De Lena M, et al. Adjuvant vinorelbine plus cisplatin versus observation in patients with completely resected stage IB-IIIA non-small-cell lung cancer (Adjuvant Navelbine International Trialist Association [ANITA]): a randomised controlled trial. *Lancet Oncol* 2006;7:719–727.
14. Pignon JP, Tribodet H, Scagliotti GV, et al; LACE Collaborative Group. Lung adjuvant cisplatin evaluation: a pooled analysis by the LACE Collaborative Group. *J Clin Oncol* 2008;26:3552–3559.
15. Douillard JY. Adjuvant chemotherapy for non-small-cell lung cancer: it does not always fade with time. *J Clin Oncol* 2010;28:3–5.
16. Hu Q, Li X, Su C, et al. Correlation between thymidylate synthase gene polymorphisms and efficacy of pemetrexed in advanced non-small cell lung cancer. *Exp Ther Med* 2012;4:1010–1016.
17. Tiseo M, Giovannetti E, Tibaldi C, et al. Pharmacogenetic study of patients with advanced non-small cell lung cancer (NSCLC) treated with second-line pemetrexed or pemetrexed-carboplatin. *Lung Cancer* 2012;78:92–99.
18. Smit EF, Burgers SA, Biesma B, et al. Randomized phase II and pharmacogenetic study of pemetrexed compared with pemetrexed plus carboplatin in pretreated patients with advanced non-small-cell lung cancer. *J Clin Oncol* 2009;27:2038–2045.
19. Adjei AA, Salavaggione OE, Mandrekar SJ, et al. Correlation between polymorphisms of the reduced folate carrier gene (SLC19A1) and survival after pemetrexed-based therapy in non-small cell lung cancer: a North Central Cancer Treatment Group-based exploratory study. *J Thorac Oncol* 2010;5:1346–1353.
20. Adjei AA, Mandrekar SJ, Dy GK, et al. Phase II trial of pemetrexed plus bevacizumab for second-line therapy of patients with advanced

non-small-cell lung cancer: NCCTG and SWOG study N0426. *J Clin Oncol* 2010;28:614–619.
21. Takehara A, Kawakami K, Ohta N, et al. Prognostic significance of the polymorphisms in thymidylate synthase and methylenetetrahydrofolate reductase gene in lung cancer. *Anticancer Res* 2005;25:4455–4461.
22. Scagliotti G, Hanna N, Fossella F, et al. The differential efficacy of pemetrexed according to NSCLC histology: a review of two phase III studies. *Oncologist* 2009;14:253–263.
23. Scagliotti GV, Parikh P, von Pawel J, et al. Phase III study comparing cisplatin plus gemcitabine with cisplatin plus pemetrexed in chemotherapy-naive patients with advanced-stage non-small-cell lung cancer. *J Clin Oncol* 2008;26:3543–3551.
24. Martini N. Mediastinal lymph node dissection for lung cancer. The Memorial experience. *Chest Surg Clin N Am* 1995;5:189–203.
25. http://clinicaltrials.gov. Accessed December 11, 2013.
26. http://clinicaltrials.gov/show/NCT00389688. Accessed December 11, 2013.
27. Scagliotti GV, Pastorino U, Vansteenkiste JF, et al. Randomized phase III study of surgery alone or surgery plus preoperative cisplatin and gemcitabine in stages IB to IIIA non-small-cell lung cancer. *J Clin Oncol* 2012;30:172–178.
28. Felip E, Rosell R, Maestre JA, et al; Spanish Lung Cancer Group. Preoperative chemotherapy plus surgery versus surgery plus adjuvant chemotherapy versus surgery alone in early-stage non-small-cell lung cancer. *J Clin Oncol* 2010;28:3138–3145.
29. Gilligan D, Nicolson M, Smith I, et al. Preoperative chemotherapy in patients with resectable non-small cell lung cancer: results of the MRC LU22/NVALT 2/EORTC 08012 multicentre randomised trial and update of systematic review. *Lancet* 2007;369:1929–1937.
30. Pisters KM, Vallières E, Crowley JJ, et al. Surgery with or without preoperative paclitaxel and carboplatin in early-stage non-small-cell lung cancer: Southwest Oncology Group Trial S9900, an intergroup, randomized, phase III trial. *J Clin Oncol* 2010;28:1843–1849.
31. Martin J, Ginsberg RJ, Venkatraman ES, et al. Long-term results of combined-modality therapy in resectable non-small-cell lung cancer. *J Clin Oncol* 2002;20:1989–1995.
32. Ahmed S, Birnbaum AE, Safran HP, et al. Pathologic response after neoadjuvant carboplatin and weekly paclitaxel for early-stage lung cancer: a Brown University oncology group phase II study. *J Thorac Oncol* 2011;6:1432–1434.
33. Betticher DC, Hsu Schmitz SF, Tötsch M, et al. Mediastinal lymph node clearance after docetaxel-cisplatin neoadjuvant chemotherapy is prognostic of survival in patients with stage IIIA pN2 non-small-cell lung cancer: a multicenter phase II trial. *J Clin Oncol* 2003;21:1752–1759.
34. Ramnath N, Sommers E, Robinson L, et al. Phase II study of neoadjuvant chemotherapy with gemcitabine and vinorelbine in resectable non-small cell lung cancer. *Chest* 2005;128:3467–3474.
35. Hou J, Lambers M, den Hamer B, et al. Expression profiling-based subtyping identifies novel non-small cell lung cancer subgroups and implicates putative resistance to pemetrexed therapy. *J Thorac Oncol* 2012;7:105–114.
36. Bueno R, Richards WG, Swanson SJ, et al. Nodal stage after induction therapy for stage IIIA lung cancer determines patient survival. *Ann Thorac Surg* 2000;70:1826–1831.
37. Higgins KA, Chino JP, Ready N, et al. Persistent N2 disease after neoadjuvant chemotherapy for non-small-cell lung cancer. *J Thorac Cardiovasc Surg* 2011;142:1175–1179.
38. Stefani A, Alifano M, Bobbio A, et al. Which patients should be operated on after induction chemotherapy for N2 non-small cell lung cancer? Analysis of a 7-year experience in 175 patients. *J Thorac Cardiovasc Surg* 2010;140:356–363.
39. Smit EF, Socinski MA, Mullaney BP, et al. Biomarker analysis in a phase III study of pemetrexed-carboplatin versus etoposide-carboplatin in chemonaive patients with extensive-stage small-cell lung cancer. *Ann Oncol* 2012;23:1723–1729.
40. Peters EJ, Kraja AT, Lin SJ, et al. Association of thymidylate synthase variants with 5-fluorouracil cytotoxicity. *Pharmacogenet Genomics* 2009;19:399–401.